Unusual variants of subepidermal autoimmune bullous diseases

Buijsrogge, Jacqueline Johanna Angela

2

Inflammatory epidermolysis bullosa acquisita with coexistent IgA antibodies to plectin

Jacqueline JA Buijsrogge, Marcelus CJM de Jong, Marcel F Jonkman, and Hendri H Pas

Center for Blistering Diseases, Department of Dermatology,
University Medical Center Groningen, University of Groningen,
Groningen, the Netherlands

Published in Clin Exp Dermatol 2005; 30(5):531-4

ABSTRACT

We present a case of inflammatory epidermolysis bullosa acquisita (EBA) with IgA antibodies to plectin. Analysis of lesional skin biopsies by electron microscopy revealed the split level to be in the sublamina densa zone, fitting with the diagnosis of EBA. Direct immunofluorescence of perilesional skin demonstrated u-serrated depositions of IgG and IgA that under immunoelectron microscopy were shown to be located in the sublamina densa. In contrast, indirect immunofluorescence on salt-split skin revealed circulating IgA antibodies that stained the roof rather than the floor of the blister. Immunoblotting showed these serum antibodies to be directed to the cytoplasmic hemidesmosomal antigen plectin. The anti-plectin specificity of these antibodies was confirmed by 'knock-out' immunofluorescence analysis: the serum IgA did not bind to skin sections of a patient with plectin-deficient epidermolysis bullosa. To our knowledge, this case demonstrates for the first time the existence of IgA antibodies against plectin.

INTRODUCTION

Epidermolysis bullosa acquisita (EBA) is a chronic subepidermal bullous autoimmune disease with autoantibodies against type VII collagen. Two clinical variants are recognized: the classic type with trauma-induced blisters, milia and dystrophic nails, and the inflammatory type with acute blisters on an erythematous base resembling bullous pemphigoid or less commonly cicatricial pemphigoid.[1]

REPORT

Here we describe a patient with an impressive chronic blistering disease who visited our clinic from 1976 until her death in 1987. Although a 'cold' case we reopened her file as the diagnostic potential of laboratory assays has significantly increased in the intervening years. At her visit to us, the patient was 70 years old and suffering from severe itch of 1 year's duration. In 1970 she had undergone a total breast amputation because of adenocarcinoma, but no signs of metastasis were found during follow up. She was being treated with propranolol for hypertension.

Physical examination showed annular erythematous macules, papules, vesicles and bullae at the wrists, trunk and lower legs. Milia were present and some blisters had healed with scarring. One year after referral the mucous membranes of the oral cavity also became involved (figure 1). Treatment with prednisolone 25 mg daily showed a good response initially; however after tapering the dosage the symptoms relapsed. From 1977 to 1987, treatment was attempted with 50-100 mg dapsone daily for years, 50-100 mg azathioprine daily for 4 years, and systemic and topical steroids, but with little or no durable effect. Until her death in 1987 from a cardiac infarction, the patient continued to have the itchy blistering disease, hypertension, and angina pectoris.

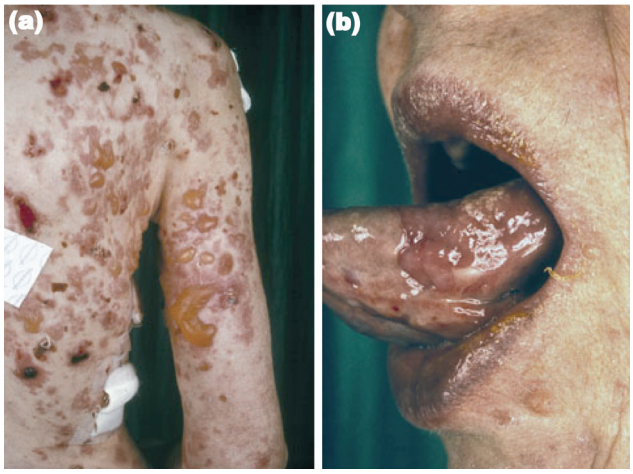

Figure 1. (a) Tense vesicles and bullae with serous content and erosions on figurate erythematous patches and urticarial plaques. (b) Erosions on the tongue.

Histopathology of lesional skin showed an infiltrate-poor subepidermal blister, with a few neutrophilic granulocytes in proteineous blister fluid. Acantholysis was absent. Subpapillary oedema and granulocytes in the dermal papillae were occasionally seen. Electron microscopy revealed the cleavage level to be in the sublamina densa zone (figure 2a). Direct immunofluorescence (IF) of perilesional skin showed linear staining of IgA and IgG along the basement membrane zone (BMZ). At first the IgA dominated, but later on IgG became more prominent. Both depositions were of the u-serrated type, recently described as indicating antibodies targeting type VII collagen (figure 2b).[2] Immunoelectron microscopy on frozen sections showed the IgA and IgG depositions to be confined to the sublamina densa zone (figure 2c).[3]

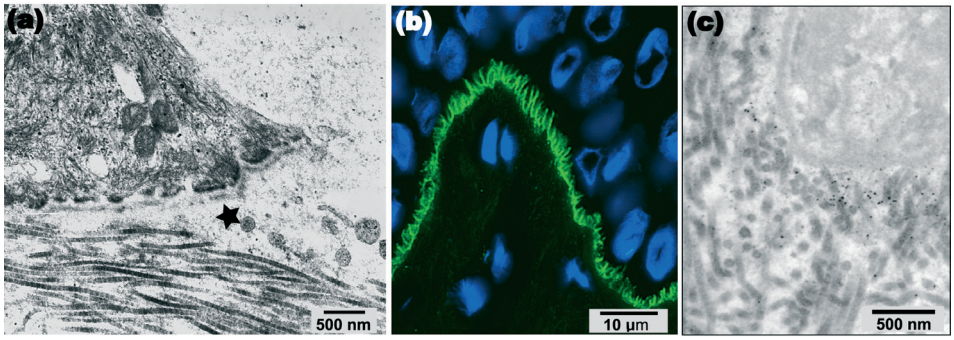

Figure 2. Skin biopsy analysis indicates epidermolysis bullosa acquisita.
(a) Electron microscopy of lesional skin reveals separation (*) in the sublamina densa zone. (b) Direct immunofluorescence analysis of perilesional skin shows an u-serrated pattern of IgA deposition along the basement membrane.

Serum IF for BMZ antibodies performed on monkey oesophagus substrate had always been negative. Repeat testing on the more sensitive salt-split skin substrate showed that the IgA surprisingly labelled the roof of the split (titre 1/10). Immunoblotting showed circulating antiplectin IgA antibodies (figure 3a) with no reactivity against other antigens, including type VII collagen.

To verify that these immunoblot binding antibodies could also bind to skin, we eluted the IgA from the blot, and using IF microscopy showed that on a section of normal human skin, it bound in a pattern corresponding with the distribution of plectin, i.e. along the BMZ and around the cell borders of keratinocytes (not shown).

Plectin is a member of the plakin protein family and is expressed in a variety of cell types including muscle and epithelial cells. It mediates the network formation of intermediate filaments, the interlinking these to microtubules and microfilaments, and anchoring them to the plasma and nuclear membrane. Absence of plectin results in epidermolysis bullosa simplex with muscle dystrophy (EBS-MD).[4]

Plectin-deficient EBS-MD skin was used as IF substrate for further proof that the serum IgA was directed to plectin. Where the patient's IgA bound the BMZ of normal skin, no binding was seen to plectin-deficient skin (figures 3b,c).

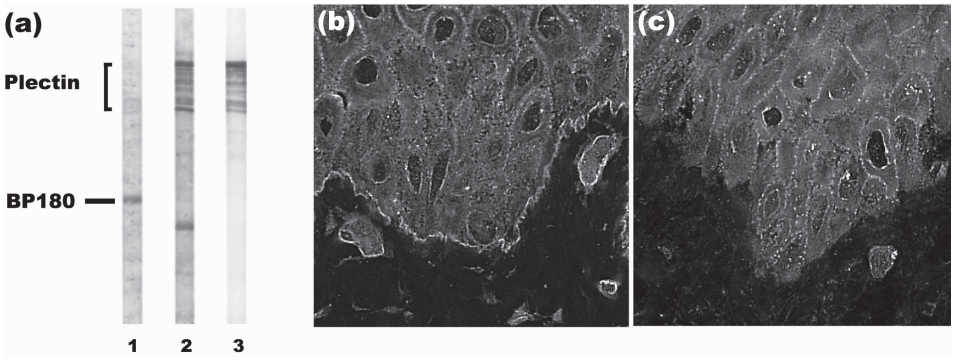

Figure 3. Serum IgA antibodies are directed to plectin.
(a) Immunoblot analysis of patient serum shows IgA binding to several high molecular weight protein bands (lane 2) completely identical with the binding pattern of anti-plectin monoclonal antibody HD-121 (lane 3).[13] The additional lower band seen with the patient serum was non-specific as it did not bind to skin after affinity elution. Lane 1 shows a control serum of a LAD patient with IgA antibodies against BP180. (b) Patient serum IgA binds to the BMZ of normal human skin, but does not bind to plectin-deficient EBS skin (c).

The final diagnosis of inflammatory EBA was based on the clinical picture in combination with the split level, the sublamina densa deposition and the u-serrated IF pattern. The serum results seemingly contrasted with the EBA diagnosis. In salt-split skin analysis, a roof staining was visible, which by immunoblot, affinity-purification and 'knock-out' IF analysis was demonstrated to be caused by anti-plectin antibodies. Apparently the anti-type VII collagen antibodies, which deposited *in vivo*, were of such low titre that they could not be detected by salt-split analysis. This is common in around half of EBA patients, for whom, despite clear skin immunodepositions, no circulating antibodies were found.[5] That the roof-binding IgA antibodies were directed to plectin was surprising.

To date, reports on anti-plectin autoantibodies are rare. So far only one case has been observed and this clinically resembled bullous pemphigoid.[6] In paraneoplastic pemphigus (PNP), the incidence of anti-plectin IgG appears higher, as Proby *et al.* found 13 cases with anti-plectin IgG among 16 PNP patients.[7] IgA to plectin, as in our patient, has not been described previously and the pathological significance of our finding is not yet clear. As the blister was formed in the sublamina densa zone the anti-type VII collagen antibodies in this particular case appear more pathogenic than the IgA to plectin.

In perilesional skin we did not detect deposition of IgA at the inner hemidesmosomal plaque, and likewise we found no n-serrated pattern in direct IF, as would have been expected if deposition had occurred at the hemidesmosome.[2] This indicates that plectin antibodies cannot

reach their intracellularly situated target in intact keratinocytes, but cell damage allows entry to the antigen. Nevertheless, the rare combination of antibody specificities may have contributed to the severe clinical pattern. Two inflammatory EBA cases with additional antibodies have been described previously, with accompagnying IgG against, respectively, laminin-332 and BP180.[8,9] These dual responses may reflect epitope spreading, a process by which, owing to tissue damage from an autoimmune inflammatory process, exposure of a previously 'sequestered' antigen leads to a secondary autoimmune response to this new antigen.[10]

ACKNOWLEDGEMENTS

We wish to thank Guus Kloosterhuis for expert technical assistance and Dr K. Owaribe (Nagoya, Japan) for supplying monoclonal antibody HD-121.

REFERENCES

1. Gammon WR, Briggaman RA, Woodley DT, et al. Epidermolysis bullosa acquisita--a pemphigoid-like disease. J Am Acad Dermatol 1984; 11:820-32.

2. Vodegel RM, Jonkman MF, Pas HH, de Jong MC. U-serrated immunodeposition pattern differentiates type VII collagen targeting bullous diseases from other subepidermal bullous autoimmune diseases. Br J Dermatol 2004; 151:112-18.

3. Prost C, Dubertret L, Fosse M et al. A routine immunoelectron microscopic technique for localizing an autoantibody on epidermal basement membrane. Br J Dermatol 1984; 110:1-7.

4. McLean WH, Pulkkinen L, Smith FJ, Rugg EL, Lane EB, Bullrich F et al. Loss of plectin causes epidermolysis bullosa with muscular dystrophy: cDNA cloning and genomic organization. Genes Dev 1996; 10:1724-35.

5. Gammon WR, Briggaman RA. Epidermolysis bullosa acquisita and bullous systemic lupus erythematosus. Diseases of autoimmunity to type VII collagen. Dermatol Clin 1993; 11:535-47.

6. Laffitte E, Favre B, Fontao L, Riou S, Jaunin F, Tamm K et al. Plectin, an unusual target antigen in bullous pemphigoid. Br J Dermatol 2001; 144:136-8.

7. Proby C, Fujii Y, Owaribe K, Nishikawa T, Amagai M. Human autoantibodies against HD1/plectin in paraneoplastic pemphigus. J Invest Dermatol 1999; 112:153-6.

8. Jonkman MF, Schuur J, Dijk F, Heeres K, de Jong MC, van der Meer JB et al. Inflammatory variant of epidermolysis bullosa acquisita with IgG autoantibodies against type VII collagen and laminin alpha3. Arch Dermatol 2000; 136:227-31.

9. Kawachi Y, Ikegami M, Hashimoto T, Matsumura K, Tanaka T, Otsuka F. Autoantibodies to bullous pemphigoid and epidermolysis bullosa acquisita antigens in an infant. Br J Dermatol 1996; 135:443-7.

10. Chan LS, Vanderlugt CJ, Hashimoto T et al. Epitope spreading: lessons from autoimmune skin diseases. J Invest Dermatol 1998; 110:103-9.